Resveratrol and obesity: Can resveratrol relieve metabolic disturbances?

Biochimica et Biophysica Acta 1852 (2015) 1137–1144

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbadis

Review

Resveratrol and obesity: Can resveratrol relieve metabolic disturbances?☆

Marlies de Ligt, Silvie Timmers, Patrick Schrauwen *

Department of Human Biology, NUTRIM School for Nutrition, Toxicology and Metabolism, Maastricht University Medical Center, 6200MD Maastricht, The Netherlands

ARTICLE INFO

Article history:
Received 22 August 2014
Received in revised form 31 October 2014
Accepted 11 November 2014
Available online 18 November 2014

Keywords:
Resveratrol
Sirtuin
Obesity
Type 2 diabetes
Mitochondria

ABSTRACT

There is an increasing need for novel preventive and therapeutic strategies to combat obesity and related metabolic disorders. In this respect, the natural polyphenol resveratrol has attracted significant interest. Animal studies indicate that resveratrol mimics the effects of calorie restriction via activation of sirtuin 1 (SIRT1). SIRT1 is an important player in the regulation of cellular energy homeostasis and mitochondrial biogenesis. Rodent studies have shown beneficial effects of resveratrol supplementation on mitochondrial function, glucose metabolism, body composition and liver fat accumulation. However, confirmation of these beneficial effects in humans by placebo-controlled clinical trials remains relatively limited. This review will give an overview of pre-clinical and clinical studies examining the effects of resveratrol on obesity-induced negative health outcomes. This article is part of a Special Issue entitled: Resveratrol: Challenges in translating pre-clinical findings to improved patient outcomes.

1. Introduction

The prevalence of obesity is increasing tremendously worldwide. A recent systematic analysis for the Global Burden of Disease Study reported a worldwide increase in overweight and obesity between 1980 and 2013 from 28.8% to 36.9% in men and 29.8% to 38.0% in women [1]. Obesity presents a health risk, partly due to ectopic fat accumulation; fat accumulation in non-adipose tissue such as liver and skeletal muscle. Accumulation of fat in the liver, when unrelated to alcohol intake, is a strong independent marker of dyslipidaemia and insulin resistance. Insulin resistance in turn predisposes to the development of type 2 diabetes (T2D) [2–4]. Impaired mitochondrial function is also often seen in obese and/or T2D patients [5]. Mitochondria play a central role in energy homeostasis and substrate metabolism. Therefore, reduced mitochondrial function has substantial effects on glucose and lipid metabolism, deteriorating metabolic health.

The rise in obesity prevalence is predominantly caused by changes in lifestyle, such as decreased physical activity and increased intake of energy-dense food. The primary solution for this obesity epidemic, and its related negative effects on public health, should therefore also be sought in changing lifestyle. Increasing the amount of physical activity or decreasing energy intake are proven effective therapeutic strategies to positively influence health outcomes related to obesity. Correspondingly, restricting calorie intake for six months leads to an improvement in insulin sensitivity [6], which in turn is accompanied by an increase in muscle mitochondrial biogenesis [7]. However, people in general have difficulties following strict exercise training or dieting regimes. Alternative treatments are therefore highly sought after. This has led to the search for compounds that can initiate beneficial health effects similar to those from exercise training or calorie restriction.

2. Resveratrol

Resveratrol (3, 5, 4′ trihydroxystilbene) is a polyphenol naturally present in and produced by several plants. The richest source of natural resveratrol is *Polygonum cuspidatum*, a plant known from traditional Chinese and Japanese medicine [8]. Smaller amounts of resveratrol can also be found in peanuts, grapes, red wine and mulberries [9]. In 2003, Howitz et al. [9] identified resveratrol as a small-molecule activator of sirtuin 1 (SIRT1). SIRT1, like all members of the sirtuin family, requires nicotinamide adenine dinucleotide (NAD^+) for its deacetylating activity [10]. The dependence of SIRT1 on NAD^+ strongly links its activity to cellular energy levels. SIRT1 is induced both by calorie restriction and exercise [11] and plays an important role in the regulation of lipid and glucose homeostasis [12]. The fact that SIRT1 is closely connected to cellular energy levels and energy homeostasis makes it an interesting molecular target for treatment of metabolic disorders such as obesity. Considering resveratrol has been identified as a small-molecule activator of SIRT1, it is not surprising that resveratrol has been said to have calorie restriction-like effects [13–16]. However, there is debate

☆ This article is part of a Special Issue entitled: Resveratrol: Challenges in translating pre-clinical findings to improved patients outcomes.

* Corresponding author at: Department of Human Biology, NUTRIM School for Nutrition, Toxicology and Metabolism, Maastricht University Medical Center, PO Box 616, 6200MD Maastricht, The Netherlands. Tel.: +31 43 388 1502; fax: +31 43 367 0976.
E-mail address: p.schrauwen@maastrichtuniversity.nl (P. Schrauwen).

http://dx.doi.org/10.1016/j.bbadis.2014.11.012
0925-4439/

whether resveratrol activates SIRT1 directly [9,17,18] or rather via activation of AMP-activated protein kinase (AMPK) [19,20]. AMPK and SIRT1 both play a crucial role in energy homeostasis and their activity is closely interrelated. It is therefore difficult to identify whether resveratrol activates SIRT1 or AMPK or both, either direct or indirect. Recently, Park et al. [21] proposed that the metabolic effects of resveratrol might result from competitive inhibition of cAMP-degrading phosphodiesterases, leading to elevated cAMP levels. Consequently, through a cascade of effects, this could lead to activation of AMPK, followed by an increase in NAD^+ and finally an increase in SIRT1 activity [21]. Unfortunately, the exact mechanism is still unknown. Despite the mechanism of action, resveratrol is a promising candidate for treatment and prevention of metabolic diseases by mimicking calorie restriction-like effects.

The aim of this review is to evaluate the potential effects of resveratrol on obesity-related health outcomes mainly in humans, both in experimental and clinical settings (see Table 1 for an overview of published peer-reviewed clinical trials on resveratrol and obesity-related health outcomes). Studies that used grape extract containing resveratrol or other formulas with multiple components are not taken into account. The effects of resveratrol on mitochondrial function, body composition, energy expenditure, insulin resistance and liver fat accumulation will be evaluated.

3. Effects of resveratrol on muscle mitochondrial function

Excessive energy intake and a low level of physical activity will lead to accumulation of fat in adipose tissue. In turn, this excessive fat accumulation can lead to lipid overflow and accumulation of fat in non-adipose tissue, such as the liver and skeletal muscle [22,23]. In general, muscle fat accumulation correlates negatively with insulin sensitivity [24] especially when mitochondrial fat oxidative capacity is low. Indeed, T2D patients and people at high risk of developing T2D are characterised by high intramyocellular lipid levels and a decreased mitochondrial fatty acid oxidative capacity [25,26]. Peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α) is a crucial regulator of mitochondrial metabolism and biogenesis, and a downstream effector of the AMPK-SIRT1 signalling pathway [27]. Several studies have reported a reduction in PGC-1α gene expression in T2D patients [28,29]. This disrupted gene expression pattern can also be seen in non-diabetic offspring of T2D patients [29]. It has been suggested that a low PGC-1α gene expression can lead to reduced generation of mitochondrial proteins, resulting in loss of mitochondrial capacity and decreased insulin sensitivity [29]. Exercise training and calorie restriction are proven effective strategies to improve muscle mitochondrial oxidative capacity, paralleled by improvements in whole body insulin sensitivity [7,30,31]. Thus, six months of calorie restriction has been shown to increase expression of AMPK, PGC-1α and SIRT1, increase mitochondrial DNA content and reduce fasting insulin levels [7]. Therefore, improving muscle oxidative capacity appears to be an effective strategy for counteracting obesity-induced insulin resistance and T2D.

A couple of animal studies have actually investigated the effects of resveratrol on muscle mitochondrial capacity. A rodent study by Lagouge et al. [32] included four different intervention groups: male C57BL/6J mice on a high-fat diet (HFD) with or without a dose of 400 mg/kg/day (mpk) of resveratrol or on a chow diet with or without resveratrol (400 mpk). The intervention period was 15 weeks. They found that mitochondria in non-oxidative muscle fibres of resveratrol-treated HFD mice were larger and denser, and mitochondrial DNA content increased compared with HFD animals that did not receive resveratrol. Um et al. [20], who also treated male C57BL/6J mice whilst on a HFD with 400 mpk resveratrol, reported increased mitochondrial content (measured by cytochrome C protein levels and mitochondrial DNA) compared with no resveratrol-treatment after an intervention period of 13 weeks. In addition, they measured a decrease in the content of the fatty acid intermediates diacylglyceride and ceramide in skeletal muscle. To further investigate mitochondrial activity, both studies measured PGC-1α expression in skeletal muscle. PGC-1α mRNA [20,32] and protein [32] significantly increased in resveratrol-treated animals. Additionally, resveratrol treatment increased physical endurance of mice, as evidenced by increased running time [20,32]. A recent study by Price et al. [17], using two different doses of resveratrol (25–30 mpk and 215–300 mpk), also found beneficial effects of resveratrol on mitochondrial biogenesis and function. The mice were fed a HFD of a standard diet for eight months. The HFD led to significantly impaired function of mitochondria isolated from skeletal muscle. Supplementation with either of the two resveratrol doses for eight months prevented the HFD-induced mitochondrial dysfunction. Thus, increases were measured compared with HFD animals without resveratrol supplementation in: ADP-stimulated respiration (state 3), FCCP-induced maximal oxidative respiration (state u), mitochondrial membrane potential, and cellular ATP levels. These levels were comparable to mice fed a standard diet. In addition, treatment with resveratrol resulted in a fibre type switch towards more oxidative muscle fibre types, and prevented the HFD-induced decline in mitochondrial content (measured by citrate synthase activity and by mitochondrial DNA content). Interestingly, when SIRT1 knockout mice were used none of the above-mentioned effects of resveratrol were observed. Moreover, SIRT1 overexpression resulted in similar effects as resveratrol treatment in wild type mice. The authors therefore concluded that SIRT1 plays a crucial role in improving mitochondrial function by resveratrol supplementation (25–30 mpk). Chen et al. [33] performed a study with male Sprague–Dawley rats fed a normal diet, HFD or HFD with resveratrol (100 mpk by intragastric administration) and found results comparable to Price et al. [17]. Hence, resveratrol-treatment increased SIRT1 activity and mitochondrial biogenesis, compared to a HFD without resveratrol. Furthermore, resveratrol reverted the decline in subsarcolemmal mitochondrial citrate synthase and electron transport chain activities and decreased IMCL content. Pearson et al. [34] investigated the effects of resveratrol on muscle mitochondrial function in a non-obese animal model. One-year old male C57BL/6NIA mice received a chow diet, both diets with and without resveratrol added (~30.9 mpk). An additional group of mice were fed every-other-day, which is a form of calorie restriction. They concluded that resveratrol shifts muscle mitochondrial gene expression patterns in mice on a standard diet towards those on a calorie restriction diet.

Together, these findings from animal studies indicate that resveratrol can influence mitochondrial biogenesis via activation of the AMPK–SIRT1–PGC-1α axis. This has led to the generally accepted idea that improving mitochondrial function by resveratrol supplementation is a promising strategy for improving metabolic health in humans. In accordance with findings from rodent studies, we have demonstrated in a previous clinical trial that resveratrol-treatment leads to activation of AMPK, increases SIRT1 and PGC-1α protein levels and increases citrate synthase activity [13]. Thus, Timmers et al. [13] studied 11 obese but otherwise healthy males receiving a dose of 150 mg resveratrol or placebo per day for 30 days, in a double-blind cross over design. No difference was found in mitochondrial content, in contrast to animal studies [20,32]. However, muscle mitochondrial fatty acid oxidative capacity was improved on a fatty acid-derived substrate (state 3 respiration), as determined by increased mitochondrial respiration. In contrast to data from animal studies, IMCL content in the vastus lateralis muscle was increased upon resveratrol-treatment. Combining the improvement in muscle fat oxidative capacity, increased IMCL content and other beneficial metabolic adaptations found in this study, the hypothesis emerged that resveratrol-treatment could have an endurance training-like effect [14,30]. One other human intervention investigated the effect of resveratrol on mitochondrial function, although indirectly by examining gene expression pathways related to mitochondrial function [15]. Thus, Yoshino et al. [15] studied non-obese postmenopausal women with normal glucose tolerance. Fifteen women received resveratrol (75 mg per day) and fourteen women received placebo, for a period of 12 weeks. Contradicting Timmers et al. [13], they concluded that

Table 1
Summary of peer-reviewed clinical trials in the field of obesity.

Author, country [ref]	Population (n)	Design	Dose of resveratrol	Duration	Objective	Outcome
Bhatt et al., India [43]	T2D men and women on oral hypoglycemic treatment (57)	Randomised, prospective, open-label control trial	250 mg once daily	3 mo	Glycemic control and associated risk factors	Resveratrol significantly improved mean HbA1c, systolic blood pressure, total cholesterol and total protein. No changes in body weight and high-density lipoprotein and low-density lipoprotein cholesterol.
Brasnyo et al., Hungary [59]	T2D men on oral glucose lowering medication (19)	Randomised, placebo-controlled, double-blind parallel design	5 mg twice daily	4 wks	Insulin sensitivity and oxidative stress	Resveratrol significantly decreased insulin resistance (measured by HOMA-IR) and urinary ortho-tyrosine excretion (as a measure of oxidative stress), whilst it increased the pAkt:Akt ratio in platelets.
Chachay et al., Australia [55]	Overweight/obese men with NAFLD (20)	Randomised, placebo-controlled parallel design	3000 mg once daily	8 wks	Insulin resistance, hepatic steatosis and abdominal fat distribution	Resveratrol did not reduce insulin resistance (measured by a hyperinsulinemic euglycemic clamp), steatosis, or abdominal fat distribution. No change was observed in plasma lipids or antioxidant activity. Levels of ALT and AST increased significantly among patients in the resveratrol group until week 6
Crandall et al., USA [44]	Elderly men and women with impaired glucose tolerance (10)	Open-label study	1, 1.5 or 2 g once daily	4 wks	Glucose metabolism and vascular function	Resveratrol (1.5 and 2 g) decreased peak glucose and 3-h glucose AUC following a meal, and improved Matsuda Index for insulin sensitivity. Weight, blood pressure and lipids were unchanged. There was a trend towards improved postmeal reactive hyperemia index
Dash et al., Canada [56]	Overweight/obese men with mild hypertriglycemia (8)	Randomised, placebo-controlled, double-blind crossover study	1000 mg daily the first week, 2000 mg daily the second week	2 wks	Intestinal and hepatic lipoprotein turnover	Resveratrol did not significantly affect insulin sensitivity (measured by HOMA-IR), fasting or fed plasma triglyceride concentration. Reduction apoB-48 production without significant effect on fractional catabolic rate. Resveratrol reduced apoB-100 and fractional catabolic rate.
Elliott et al. [58]	T2D patients		2.5 or 5 g once daily	28 d	Insulin sensitivity	Resveratrol (5 g) decreased fasting and postprandial glucose and insulin
Magyar et al., Hungary [57]	Male and female patients with stable coronary artery disease (40)	Randomised, placebo-controlled, double-blind trial	10 mg once daily	3 mo	Cardioprotective effects	Resveratrol improved left ventricle diastolic function, endothelial function, lowered LDL-cholesterol level and protected against unfavourable haemorheological changes. No significant difference in HbA1c.
Movahed et al., Iran [46]	T2D men and women on oral hypoglycemic treatment (66)	Randomised, placebo-controlled, double-blinded parallel clinical trial	500 mg twice daily	45 d	Antihyperglycemic effects	Resveratrol significantly decreased systolic blood pressure, fasting blood glucose, HbA1c, insulin, and insulin resistance (measured by HOMA-IR), whilst HDL was significantly increased. Liver and kidney function markers were unchanged
Poulsen et al., Denmark [45]	Healthy obese men (24)	Randomised, placebo-controlled, double-blind parallel design. Resveratrol	500 mg thrice daily	4 wks	Metabolic effects	Resveratrol did not change insulin sensitivity (measured by a hyperinsulinemic euglycemic clamp), blood pressure, resting energy expenditure, lipid oxidation rates, ectopic or visceral fat content, or inflammatory or metabolic biomarkers
Timmers et al., The Netherlands [13]	Healthy obese men (11)	Randomised, placebo-controlled, double-blind crossover design	75 mg twice daily	30 d	Metabolic effects	Resveratrol improved metabolic profile: resveratrol reduced sleeping and resting metabolic rate. In muscle resveratrol activated the AMPK–SIRT1–PGC1α axis. A reduction in blood glucose and insulin levels and liver. Improved muscle mitochondrial function and reduced inflammation markers in the blood.
Yoshino et al., USA [15]	Nonobese postmenopausal women with normal glucose tolerance (29)	Randomised, placebo-controlled, double-blind parallel design	75 mg once daily	12 wks	Metabolic effects	Resveratrol did not change resting metabolic rate, body composition, inflammatory markers or plasma lipids. Resveratrol did not increase liver, skeletal muscle, or adipose tissue insulin sensitivity (measured by a hyperinsulinemic euglycemic clamp). Resveratrol did not affect AMPK, SIRT1, NAMPT, or PGC1α in either skeletal muscle or adipose tissue.

ref, reference; T2D, type 2 diabetes mellitus; wks, weeks; mo, months; d, days; NAFLD, non-alcoholic fatty liver disease.

resveratrol did not have an effect on the phosphorylation of AMPK or SIRT1 gene expression in skeletal muscle. It must be noted that the dose used by Timmers et al. [13] was double the dose that Yoshino et al. [15] used. Furthermore, our trial included obese men, implying that resveratrol could have beneficial effects on people with compromised health but not on healthy non-obese humans. Unfortunately, no other data from human interventions are currently available regarding the effect of resveratrol on muscle mitochondrial function.

4. Effect of resveratrol on body composition and energy expenditure

Rodent studies suggest that high doses of resveratrol (~400 mpk) can lead to a reduction in weight gain when animals are fed a HFD [32,35]. Two rodent studies by Lagouge et al. [32] and Kim et al. [35] both used a similar set-up with male C57BL/BJ mice and a resveratrol dose of 400 mpk. The study by Kim et al. [35] included three different mice groups: HFD with or without resveratrol or chow diet without resveratrol, with an intervention period of 10 weeks. Both studies were consistent in reporting a blunting of body mass gain in the HFD–resveratrol group compared with the HFD group. The blunted weight gain was accounted for by a decrease in visceral fat-pad weights and smaller adipocytes in epididymal adipose tissue [32,35]. The beneficial effect of resveratrol on body mass and composition could not be attributed to decreased food intake, since both resveratrol-treated and non-resveratrol-treated animals consumed an equal amount of calories. Lagouge et al. [32] measured energy expenditure of the mice by indirect calorimetry and recorded a higher basal energy expenditure (EE) accompanied by a decrease in locomotor activity. This suggests that resveratrol had a stimulating effect on energy expenditure. The latter results are in accordance with those found in a non-human primate model of obesity [36]. Six male grey mouse lemurs received a dose of ~200 mpk resveratrol for four weeks, during their winter body-mass gain period. Supplementation of resveratrol led to an increase in resting EE of 29% and a reduction in body-mass gain, compared with the control period. Contrary to the rodent studies, the grey mouse lemurs decreased their food intake upon resveratrol-treatment, thereby contributing to the decrease in weight gain. As a follow-up of this short-term non-human primate study a larger group of animals received the supplements (~200 mpk) for one year [37]. Overall, there were no differences in body weight, food intake or physical activity between resveratrol and control animals. Yet, at the beginning of the so-called long day period (spring/summer period) an increase in fat free mass was noted in the resveratrol-supplemented lemurs, which was accompanied by an increase in total daily EE and resting EE. Overall, the findings from animal studies suggest that resveratrol could stimulate energy expenditure and protect against HFD-induced weight gain.

To further examine how resveratrol might increase energy expenditure, Lagouge et al. [32] examined thermogenesis during a cold test with the focus on brown adipose tissue (BAT). In mice the main contributor to heat production is BAT, which in contrast to white adipose tissue stores little fat and burns fat to produce heat and regulate body temperature; non-shivering thermogenesis [38]. It was found that mitochondria were significantly larger and also mitochondrial DNA content was increased in BAT of resveratrol-treated mice [32]. These changes were accompanied by an increase in gene expression of SIRT1, a decrease in PGC-1α acetylation and an increase in PGC-1α activity. These findings suggest that resveratrol may affect BAT metabolism. Accordingly, a significant increase in uncoupling protein 1 (UCP1) and SIRT1 gene expression in BAT was found in male mice treated with resveratrol for eight weeks (400 mpk) [39] and in Sprague–Dawley rats treated for six weeks (30 mpk) [40]. Also, gene expression of PGC-1α and peroxisome proliferator-activated receptor β/δ (PPARβ/δ) was significantly higher in BAT of the resveratrol-supplemented rats. Bone Morphogenetic Protein 7 (BMP7) could also be involved in the effect of resveratrol on BAT, since BMP7 is a crucial factor in brown adipogenesis and 2 months of resveratrol-treatment (400 mpk) were shown to increase BMP7 expression in BAT of mice [39]. Previous studies demonstrated that BMP7 promotes brown pre-adipocyte differentiation [41]. In vitro treatment of brown pre-adipocytes with BMP7 induced PGC-1α and increased gene expression of UCP1 and PPARγ [41]. In addition, when gene expression of BMP7 was stimulated in vivo, this resulted in a significant increase in BAT mass accompanied by an increase in energy expenditure and a reduction in weight gain [41]. Taken together, the data illustrate that resveratrol, via activation of SIRT1, UCP1 and potentially BMP7, could influence brown adipocyte differentiation. This in turn could lead to increased energy expenditure and eventually weight loss. Whether similar effects of resveratrol are present in humans unfortunately remains unclear. It is difficult to investigate molecular aspects of BAT metabolism in humans since BAT is located in regions of the human body that are difficult to reach. However, future human studies could investigate the effect of resveratrol on BAT activity using PET-CT scanning [42].

So far, no effect of resveratrol has been found in human trials concerning body weight [13,15,43–46] or body composition [15,45]. It is however important to note that in the human studies performed so far, participants were generally instructed to consume their usual diet and were not challenged with a HFD or high-calorie diet (HCD) in contrast to the animal studies. Moreover, in human interventions, relatively low doses of resveratrol have been used ranging from 75 to 2000 mg/d. In that respect, it is interesting to mention that also in rodents a relatively low dose of resveratrol (~22.4 mpk) does not lead to a decrease in body weight [19]. In addition, the possibility cannot be excluded that metabolism of resveratrol differs between humans and animals. The duration of the resveratrol supplementation could also have been too short in the human intervention studies performed so far. Strikingly, we have previously demonstrated that 30 days of resveratrol supplementation (150 mg/day) leads to a *reduction* in energy expenditure in healthy obese males [13]. Thus, in sharp contrast to animal studies, resveratrol supplementation resulted in a lower sleeping metabolic rate as well as a lower postprandial energy expenditure [13]. Long-term studies in humans are needed to provide definitive answers as to whether resveratrol can affect energy expenditure or body composition in humans.

5. Effects of resveratrol on insulin sensitivity

Obesity is one of the major determinants of insulin resistance and T2D. As described earlier in this review, improving muscle oxidative capacity can counteract obesity-induced insulin resistance [30]. Rodent studies have demonstrated that resveratrol can have beneficial effects on glucose homeostasis in animal models of obesity, diabetes and metabolic dysfunction [19,20,32,35,47–52]. These studies used variable doses of resveratrol, ranging from 2.5 to 400 mpk. In addition, the exposition time also varied from between 2 weeks and 16 months. Lagouge et al. [32] investigated mice on a HFD with or without resveratrol supplementation (400 mpk) for 15 weeks. A significant reduction in fasting insulin levels was found in resveratrol-treated animals, compared with HFD animals without resveratrol supplementation. This decrease in insulin was not accompanied by alterations in fasting glucose levels. In addition, insulin sensitivity was assessed by the hyperinsulinemic euglycemic clamp technique, which is regarded as the gold standard for measuring insulin sensitivity. Improved insulin sensitivity was found in the resveratrol group compared with the HFD group without resveratrol. Shang et al. [48] also used the hyperinsulinemic euglycemic clamp technique to determine insulin sensitivity. Resveratrol treatment (100 mpk for 10 weeks) in HFD-fed male Wistar rats resulted in a higher glucose infusion rate accompanied by reduced fasting plasma insulin levels, compared with HFD animals that did not receive resveratrol. Sun et al. [52] used a relatively low dose of resveratrol, namely 2.5 mpk for 16 weeks, and found that resveratrol treatment significantly improved glucose tolerance (as determined by a glucose tolerance test) in HFD-fed male C57BL/6J mice with glucose disposal

curves becoming comparable to mice fed a chow diet. In the same study, resveratrol administration also considerably improved insulin sensitivity as determined by an insulin tolerance test. These findings indicate that resveratrol has the potency to improve insulin sensitivity in diet-induced obesity in rodents, and that these effects are independent of dose and exposition time. The effect of resveratrol on insulin sensitivity has also been tested on non-human primates. Hence, Marchal et al. [53] performed a study with grey mouse lemurs fed a standard diet with or without resveratrol (200 mpk). Insulin sensitivity was determined with the oral glucose tolerance test and the homeostasis model assessment of insulin resistance (HOMA-IR index). They concluded that 33 months of resveratrol supplementation improved glucose tolerance and thereby affected insulin sensitivity without changes in basal insulin secretion. Jimenez-Gomez et al. [54] used male adult rhesus monkeys in their 2-year trial. Animals were randomised in one of three groups: high-fat high-sugar diet (HFS) with resveratrol (n = 10), HFS without resveratrol (n = 10) or a standard diet (n = 4). The dose of resveratrol was 40 mg twice a day for the first year, and 240 mg twice a day during the second intervention year. Resveratrol supplementation for two years increased SIRT1 protein expression, decreased adipocyte size, and improved insulin sensitivity in visceral but not subcutaneous white adipose tissue. Regarding insulin sensitivity, an increase of insulin receptor substrate 1 protein levels, a decrease of Akt serine 473 phosphorylation, and an increase in the content of insulin-responsive GLUT4 glucose transporter was measured in visceral fat depots from fasted rhesus monkeys on a HFS diet supplemented with resveratrol as compared with animals on a HFS diet without resveratrol supplementation. These findings from animal studies led to the hypothesis that resveratrol could be a promising anti-diabetic agent for humans.

So far, eleven clinical trials have investigated the effects of resveratrol on glucose homeostasis and insulin sensitivity. Three of these studies used the gold-standard hyperinsulinemic euglycemic clamp technique to determine insulin sensitivity [15,45,55]. Thus, Poulsen et al. [45] supplemented healthy obese men, in a parallel-group design, with a daily dose of 1500 mg resveratrol (n = 12) or placebo (n = 12) for four weeks. The subjects consumed the resveratrol supplements in doses of 500 mg three times a day. Insulin sensitivity was measured by hyperinsulinemic euglycemic clamp in conjunction with ^{3}H-labelled glucose tracer infusion. No changes upon resveratrol supplementation were observed in endogenous glucose production, oxidative glucose disposal or non-oxidative glucose disposal. Yoshino et al. [15] also did not find a difference in insulin sensitivity in non-obese postmenopausal women who received 75 mg resveratrol per day. Twelve weeks of resveratrol supplementation did not induce changes in liver, skeletal muscle or adipose tissue insulin sensitivity, as measured by the hyperinsulinemic euglycemic clamp in combination with stable isotope labelled tracer infusion. The third study, by Chachay et al. [55], included 20 overweight or obese men diagnosed with non-alcoholic fatty liver disease (NAFLD). The participants were randomly assigned to groups given 3000 mg of resveratrol (n = 10) or placebo (n = 10) daily for eight weeks. Also in this study resveratrol administration did not improve insulin sensitivity, as measured by the clamp technique.

The studies that used other measures of glucose homeostasis or insulin sensitivity are also inconsistent in effects of resveratrol. Timmers et al. [13] did find positive metabolic changes in healthy obese men (n = 11) supplemented with resveratrol. Thirty days of resveratrol supplementation significantly reduced blood glucose and insulin levels and improved the HOMA-IR index compared with the placebo condition. Crandall et al. [44] found beneficial effects of resveratrol in overweight and obese men and women with impaired glucose tolerance. Ten subjects were enrolled in this 4-week open-label study. Resveratrol-treatment for four weeks, with either 1.5 or 2 g per day, led to decreased peak glucose and 3-hour glucose area under the curve following a meal. Furthermore, both 1.5 and 2 g of daily resveratrol supplementation led to an improvement of the Matsuda index, representing improved insulin sensitivity. It must be noted that this study did not include a placebo condition. Dash et al. [56], who investigated overweight or obese men with mild hypertriglyceridemia, did not observe differences in fasting plasma glucose or insulin levels. The participants (n = 8) were studied in two experimental conditions: after treatment with resveratrol and after treatment with placebo in a randomised, double-blinded, crossover design. During the resveratrol condition participants received a daily dose of 1000 mg resveratrol for one week, followed by a daily dose of 2000 mg resveratrol during the second week. No significant improvement in HOMA-IR index was observed. One study investigated the effects of resveratrol on HbA1c, in patients with stable coronary artery disease [57]. In total 26 male and 14 female patients were enrolled, receiving either 10 mg resveratrol per day (n = 20) or a placebo (n = 20) for three months. No significant change in HbA1c was found after three months of resveratrol-treatment compared with the baseline. Unfortunately systemic conversion of resveratrol was not measured, so no statements can be made as to whether the low dose used (10 mg) was high enough to evoke an observable increase in resveratrol in blood plasma. Furthermore, the patients received medical therapy including platelet aggregation inhibitors, β-blockers, ACE-inhibitors and statins, which could have influenced the efficacy of resveratrol. Contrary to the previously mentioned studies, the studies in T2D patients (without insulin treatment) have been more consistent in reporting beneficial effects of resveratrol supplementation on blood glucose levels [46,58,59], insulin levels [46,58], insulin resistance (defined by HOMA-IR index) [46,59] and HbA1c [43,46]. Hence, Elliott et al. [58] measured a decrease in fasting and postprandial glucose and insulin after a daily administration of 5 g of resveratrol for 28 days to T2D patients. Brasnyó et al. [59] performed a double-blind, placebo-controlled trial with male T2D patients supplemented with a dose of two times 5 mg resveratrol per day (n = 10) or placebo (n = 9) for four weeks. Mean changes in insulin sensitivity (defined by HOMA-IR index), blood glucose and insulin were compared between the placebo and resveratrol group. Resveratrol significantly decreased insulin sensitivity (defined by HOMA-IR index) and decreased blood glucose levels in T2D patients on standard oral glucose lowering medication. No difference was found in serum insulin levels between the resveratrol and placebo groups at any time during the trial. Bhatt et al. [43] investigated the effects of resveratrol on HbA1c in T2D patients. Fifty-seven T2D patients, males and females combined, received a daily dose of 250 mg resveratrol (n = 28) or received no interventional-treatment (n = 29). The results revealed that three months of resveratrol supplementation significantly improved average HbA1c in T2D patients on oral hypoglycemic treatment (metformin and/or glibenclamide) compared with their baseline values. A limitation of this study is the lack of a placebo group. The fourth clinical trial in T2D patients was executed by Movahed et al. [46]. A total of 64 T2D patients, consisting of both males and females on standard diabetic treatment, completed this randomised placebo-controlled double-blinded parallel clinical trial. The intervention group was supplemented with resveratrol at a dose of 500 mg twice a day (n = 33), and the control group (n = 31) received placebo tablets both for a period of 45 days. Resveratrol-treatment significantly decreased fasting blood glucose, HbA1c, insulin and insulin sensitivity (defined by HOMA-IR index), whereas in the placebo group fasting glucose levels increased slightly compared with their baseline values.

To conclude, most rodent studies show a decrease in plasma glucose [19,35,47,49,51], a decrease in plasma insulin [19,32,35,47–49] or improved insulin sensitivity/glucose tolerance [20,32,48,50,52] upon resveratrol administration. Clinical trials in T2D patients imply an anti-diabetic effect of resveratrol, but the studies in non-diabetic patients are less consistent. Studies in T2D patients investigating the effects of resveratrol on insulin sensitivity by means of the gold-standard hyperinsulinemic euglycemic clamp technique are highly demanded.

6. Effect of resveratrol on liver fat accumulation

Excessive body weight and obesity can exert negative metabolic health effects partly via accumulation of fat in the liver [60]. Indeed, the increase in obesity prevalence is accompanied by an increase in NAFLD. NAFLD itself does not explicitly evoke a risk to health but when inflammation occurs and NAFLD progresses into non-alcoholic steatohepatitis, it strongly correlates with morbidity and mortality rates [61]. It has been suggested that resveratrol can decrease liver fat accumulation through activation of AMPK and/or SIRT1. In the liver, deletion of SIRT1 leads to impaired signalling of the lipid-sensing transcription factor PPARα resulting in decreased β-oxidation [62]. In addition, knockdown of hepatic SIRT1 results in inhibition of PGC-1α leading to increased levels of hepatic free fatty acids [63]. Sterol regulatory element-binding protein 1c (SREBP-1c) is a transcription factor that regulates the expression of genes related to triglyceride and fatty acid synthesis [64,65]. SIRT1 down-regulates SREBP-1c expression in the liver, leading to a decrease in lipid synthesis and fat storage [64,65]. These findings lead to the idea that resveratrol could stimulate hepatic fatty acid oxidation through AMPK/SIRT1-mediated activation of PGC-1α and PPARα and inhibition of SREBP-1c.

Baur et al. [19] investigated resveratrol supplementation at a dose of ~22.4 mpk in one-year old male C57BL/6NIA mice. The mice received a HCD with or without resveratrol or a standard diet, for 6 months. It was found that resveratrol prevented an increase in size and weight of the liver as seen in the HCD group. In addition, histological staining showed that accumulation of large lipid droplets in the liver was present in the HCD group but not in the group on a HCD with resveratrol. The livers of the HCD resveratrol-treated mice also had substantially more mitochondria than those of HCD animals, and were not significantly different from those in the standard diet group. These findings were accompanied by increased phosphorylation of AMPK and decreased gene expression of fatty acid synthase in the liver. In a separate cohort of the one-year-old mice on the HCD, which had been treated with a dose of 186 mpk resveratrol for six weeks, acetylation of PGC-1α was significantly lower, indicating increased activity, in resveratrol-fed animals compared with the diet matched controls. Shang et al. [48] performed a rat study with comparable results. Male Wistar rats fed a HFD developed abdominal obesity, NAFLD and insulin resistance, which was considerably improved by 10 weeks of resveratrol supplementation (100 mpk). Again, resveratrol-treatment promoted phosphorylation of AMPK in the liver, which in turn suppressed SREBP-1c and fatty acid synthase gene expression. Another rat study found that a low dose of resveratrol (30 mpk for six weeks) led to a decreased accumulation of fat in the liver and activated AMPK and PGC-1α in the male Sprague–Dawley rats [66]. However, they did not find a difference in mRNA expression of SREBP-1c, PPARα, SIRT1 or PCG-1α. Poulsen et al. [67] detected that male Wistar rats treated with 100 mg resveratrol daily for eight weeks increased hepatic mitochondrial content when on a HFD. In addition, hepatic uncoupling protein 2 (UCP2) gene expression was significantly increased compared with both control and HFD-fed animals, suggesting that a resveratrol-induced increase in mitochondrial number and UCP2 gene expression may contribute to normalising liver fat content in HFD-induced liver fat accumulation. Recently, Heebøll et al. [68] further reviewed the effects of resveratrol on experimental and clinical NAFLD. They concluded that rodent studies demonstrate a consistent decrease in cholesterol accumulation [35,69–74], hepatic triglycerides [35,47,66,69,70,72,75–79] and liver weight [19,48,70,73,76,77]. Also, several other improvements in liver fat content and anatomy were reported in rodents after resveratrol supplementation [19,48–50,67,69–72,74,76–82]. For example, improvements were noted regarding steatosis, steatohepatitis and fibrosis. Taken together, resveratrol in rodents is at least partly able to prevent liver fat accumulation induced by HFD or HCD, probably via increasing fatty acid oxidation and decreasing lipogenesis. These effects could be mediated by activation of the AMPK–SIRT1 axis.

To date, only four clinical trials have been performed investigating effects of resveratrol on liver fat accumulation. One of these studies specifically focussed on patients suffering from NAFLD, whilst the other three studies involved healthy participants. In the study by Chachay et al. [55] 10 NAFLD patients received a daily dose of 3000 mg resveratrol and another group of 10 NAFLD patients received a placebo. One of the study outcomes was hepatic steatosis assessed by magnetic resonance spectroscopy (MRS) and imaging (MRI). Chachay et al. [55] concluded that eight weeks of resveratrol administration did not significantly improve any features of NAFLD, compared with placebo. Furthermore, based on increases in liver enzymes ALT and AST, they concluded that there was an increase in hepatic stress in the resveratrol group. Timmers et al. [13] used a much lower dose of resveratrol for a shorter period of time: 150 mg/d, for 30 days. The outcome of this study was that resveratrol supplementation led to a reduction in liver fat in healthy obese males, measured by MRS. In addition, a significant reduction in liver transaminases (ALT) was found suggesting beneficial effects of resveratrol on liver function. In contrast, Poulsen et al. [45], who also investigated healthy obese males, did not find effects of resveratrol (1500 mg/d) on liver fat content (measured by MRS) or liver transaminase levels after four weeks of supplementation. The fourth clinical trial, in non-obese postmenopausal women, also did not find significant differences in liver fat (measured by MRS) after 12 weeks of supplementation (75 mg/d) [15]. Taken together; although rodent studies have generally found a lowering effect of resveratrol on liver fat content, human studies have so far been less consistent. Again, differences in doses used or duration of resveratrol treatment may explain differences between human studies so far, and more clinical studies are required.

7. Discussion and future studies

The effect of resveratrol on metabolic health has received much attention in the last decade. Pre-clinical studies have revealed promising results regarding beneficial effects of resveratrol on preventing and reversing obesity-induced metabolic disturbances. Specifically, beneficial effects have been observed in rodents supplemented with resveratrol with respect to mitochondrial function, insulin sensitivity and liver fat accumulation. These effects can be attributed to activation of the AMPK–SIRT1–PGC-1α axis. Clinical trials performed so far have been less consistent. This inconsistency can most likely be explained by differences in duration and dose of resveratrol used between studies. Relative to animal studies, clinical trials used low doses of resveratrol. For example, a dose of 400 mpk is frequently used in rodents and would translate into a dose of 30 g resveratrol per day for an average human with a body weight of 75 kg. Despite using lower doses of resveratrol in human clinical trials, plasma resveratrol concentrations have been found comparable to those found in mice studies [13]. However, a direct comparison of circulating levels between animals and humans may be difficult as in vivo metabolism of resveratrol might differ between mice and humans. Human studies would be needed to determine the optimal dose of resveratrol to activate the AMPK–SIRT1 axis. In fact only few human studies so far have determined if resveratrol administration indeed resulted in activation of the AMPK–SIRT1 axis, leaving the possibility that suboptimal doses have been used so far. In addition, long-term studies in humans are needed to investigate the possibility of cumulative effects of resveratrol on metabolic health. Finally, it is important to note that the effects of resveratrol are mainly observed in animals fed a HFD or HCD diet, implying that resveratrol is particularly potent in reversing early stage metabolic disorders. The same could apply to humans, which is supported by the fact that so far only clinical trials in overweight/obese participants [13,44] or T2D patients [43,46,58,59] have found beneficial effects.

An interesting new venue for future research would be a focus on human BAT and so-called browning of white adipose tissue. In contrast to white adipose tissue, BAT stores little fat and instead burns fat to

produce heat and regulate body temperature. This feature of BAT led to the hypothesis that stimulating BAT thermogenesis could initiate weight loss [83]. A few animal studies have been performed investigating potential effects of resveratrol on BAT thermogenesis and these studies were overall encouraging, indicating enhanced mitochondrial biogenesis [32], induction of UCP1 [32,39,40] and activation of BMP7 [40] (a promoter of brown preadipocyte differentiation) in BAT. Given the interest in the physiology of human BAT, it would be interesting to investigate if resveratrol can also activate BAT in humans.

To conclude, even though data from animal studies look promising, currently the number of clinical trials is too limited to make any firm statements regarding the effects of resveratrol on obesity-induced negative health outcomes in humans. More specifically, future research should primarily focus on dose, efficacy and chronic exposure effects of resveratrol. Finally, human intervention studies are necessary focusing on the potential of resveratrol to stimulate BAT thermogenesis.

Acknowledgements

This study was supported by the Dutch Diabetes Foundation grant 2012.00.1525 to P.S.

References

[1] M. Ng, T. Fleming, M. Robinson, B. Thomson, N. Graetz, C. Margono, E.C. Mullany, S. Biryukov, C. Abbafati, S.F. Abera, et al., Global, regional, and national prevalence of overweight and obesity in children and adults during 1980–2013: a systematic analysis for the Global Burden of Disease Study 2013, Lancet 384 (2014) 766–781.
[2] S. Deivanayagam, B.S. Mohammed, B.E. Vitola, G.H. Naguib, T.H. Keshen, E.P. Kirk, S. Klein, Nonalcoholic fatty liver disease is associated with hepatic and skeletal muscle insulin resistance in overweight adolescents, Am. J. Clin. Nutr. 88 (2008) 257–262.
[3] K.M. Korenblat, E. Fabbrini, B.S. Mohammed, S. Klein, Liver, muscle, and adipose tissue insulin action is directly related to intrahepatic triglyceride content in obese subjects, Gastroenterology 134 (2008) 1369–1375.
[4] E. Fabbrini, B.S. Mohammed, F. Magkos, K.M. Korenblat, B.W. Patterson, S. Klein, Alterations in adipose tissue and hepatic lipid kinetics in obese men and women with nonalcoholic fatty liver disease, Gastroenterology 134 (2008) 424–431.
[5] J.C. Bournat, C.W. Brown, Mitochondrial dysfunction in obesity, Curr. Endocrinol. Diabetes. Obes. 17 (2010) 446–452.
[6] D.E. Larson-Meyer, B.R. Newcomer, L.K. Heilbronn, J. Volaufova, S.R. Smith, A.J. Alfonso, M. Lefevre, J.C. Rood, D.A. Williamson, E. Ravussin, et al., Effect of 6-month calorie restriction and exercise on serum and liver lipids and markers of liver function, Obesity (Silver Spring) 16 (2008) 1355–1362.
[7] A.E. Civitarese, S. Carling, L.K. Heilbronn, M.H. Hulver, B. Ukropcova, W.A. Deutsch, S.R. Smith, E. Ravussin, C.P. Team, Calorie restriction increases muscle mitochondrial biogenesis in healthy humans, PLoS Med. 4 (2007) e76.
[8] S. Nonomura, H. Kanagawa, A. Makimoto, Chemical constituents of polygonaceous plants. I. Studies on the components of Ko-J O-Kon. (*Polygonum cuspidatum* Sieb. Et Zucc.), Yakugaku Zasshi 83 (1963) 988–990.
[9] K.T. Howitz, K.J. Bitterman, H.Y. Cohen, D.W. Lamming, S. Lavu, J.G. Wood, R.E. Zipkin, P. Chung, A. Kisielewski, L.L. Zhang, et al., Small molecule activators of sirtuins extend *Saccharomyces cerevisiae* lifespan, Nature 425 (2003) 191–196.
[10] A.A. Sauve, C. Wolberger, V.L. Schramm, J.D. Boeke, The biochemistry of sirtuins, Annu. Rev. Biochem. 75 (2006) 435–465.
[11] N.B. Ruderman, X.J. Xu, L. Nelson, J.M. Cacicedo, A.K. Saha, F. Lan, Y. Ido, AMPK and SIRT1: a long-standing partnership? Am. J. Physiol. Endocrinol. Metab. 298 (2010) E751–E760.
[12] J. Yu, J. Auwerx, The role of sirtuins in the control of metabolic homeostasis, Ann. N. Y. Acad. Sci. 1173 (Suppl. 1) (2009) E10–E19.
[13] S. Timmers, E. Konings, L. Bilet, R.H. Houtkooper, T. van de Weijer, G.H. Goossens, J. Hoeks, S. van der Krieken, D. Ryu, S. Kersten, et al., Calorie restriction-like effects of 30 days of resveratrol supplementation on energy metabolism and metabolic profile in obese humans, Cell Metab. 14 (2011) 612–622.
[14] J.J. Dube, F. Amati, M. Stefanovic-Racic, F.G. Toledo, S.E. Sauers, B.H. Goodpaster, Exercise-induced alterations in intramyocellular lipids and insulin resistance: the athlete's paradox revisited, Am. J. Physiol. Endocrinol. Metab. 294 (2008) E882–E888.
[15] J. Yoshino, C. Conte, L. Fontana, B. Mittendorfer, S. Imai, K.B. Schechtman, C. Gu, I. Kunz, F. Rossi Fanelli, B.W. Patterson, et al., Resveratrol supplementation does not improve metabolic function in nonobese postmenopausal women with normal glucose tolerance, Cell Metab. 16 (2012) 658–664.
[16] C. Canto, J. Auwerx, Targeting sirtuin 1 to improve metabolism: all you need is NAD(+)? Pharmacol. Rev. 64 (2012) 166–187.
[17] N.L. Price, A.P. Gomes, A.J. Ling, F.V. Duarte, A. Martin-Montalvo, B.J. North, B. Agarwal, L. Ye, G. Ramadori, J.S. Teodoro, et al., SIRT1 is required for AMPK activation and the beneficial effects of resveratrol on mitochondrial function, Cell Metab. 15 (2012) 675–690.
[18] M. Pacholec, J.E. Bleasdale, B. Chrunyk, D. Cunningham, D. Flynn, R.S. Garofalo, D. Griffith, M. Griffor, P. Loulakis, B. Pabst, et al., SRT1720, SRT2183, SRT1460, and resveratrol are not direct activators of SIRT1, J. Biol. Chem. 285 (2010) 8340–8351.
[19] J.A. Baur, K.J. Pearson, N.L. Price, H.A. Jamieson, C. Lerin, A. Kalra, V.V. Prabhu, J.S. Allard, G. Lopez-Lluch, K. Lewis, et al., Resveratrol improves health and survival of mice on a high-calorie diet, Nature 444 (2006) 337–342.
[20] J.H. Um, S.J. Park, H. Kang, S. Yang, M. Foretz, M.W. McBurney, M.K. Kim, B. Viollet, J.H. Chung, AMP-activated protein kinase-deficient mice are resistant to the metabolic effects of resveratrol, Diabetes 59 (2010) 554–563.
[21] S.J. Park, F. Ahmad, A. Philp, K. Baar, T. Williams, H. Luo, H. Ke, H. Rehmann, R. Taussig, A.L. Brown, et al., Resveratrol ameliorates aging-related metabolic phenotypes by inhibiting cAMP phosphodiesterases, Cell 148 (2012) 421–433.
[22] O.P. Bachmann, D.B. Dahl, K. Brechtel, J. Machann, M. Haap, T. Maier, M. Loviscach, M. Stumvoll, C.D. Claussen, F. Schick, et al., Effects of intravenous and dietary lipid challenge on intramyocellular lipid content and the relation with insulin sensitivity in humans, Diabetes 50 (2001) 2579–2584.
[23] V.B. Schrauwen-Hinderling, M.E. Kooi, M.K. Hesselink, E. Moonen-Kornips, G. Schaart, K.J. Mustard, D.G. Hardie, W.H. Saris, K. Nicolay, P. Schrauwen, Intramyocellular lipid content and molecular adaptations in response to a 1-week high-fat diet, Obes. Res. 13 (2005) 2088–2094.
[24] M. Krssak, K. Falk Petersen, A. Dresner, L. DiPietro, S.M. Vogel, D.L. Rothman, M. Roden, G.I. Shulman, Intramyocellular lipid concentrations are correlated with insulin sensitivity in humans: a ^{1}H NMR spectroscopy study, Diabetologia 42 (1999) 113–116.
[25] E.E. Blaak, D.P. van Aggel-Leijssen, A.J. Wagenmakers, W.H. Saris, M.A. van Baak, Impaired oxidation of plasma-derived fatty acids in type 2 diabetic subjects during moderate-intensity exercise, Diabetes 49 (2000) 2102–2107.
[26] M. Mensink, E.E. Blaak, M.A. van Baak, A.J. Wagenmakers, W.H. Saris, Plasma free fatty acid uptake and oxidation are already diminished in subjects at high risk for developing type 2 diabetes, Diabetes 50 (2001) 2548–2554.
[27] M.M. Mihaylova, R.J. Shaw, The AMPK signalling pathway coordinates cell growth, autophagy and metabolism, Nat. Cell Biol. 13 (2011) 1016–1023.
[28] V.K. Mootha, C.M. Lindgren, K.F. Eriksson, A. Subramanian, S. Sihag, J. Lehar, P. Puigserver, E. Carlsson, M. Ridderstrale, E. Laurila, et al., PGC-1alpha-responsive genes involved in oxidative phosphorylation are coordinately downregulated in human diabetes, Nat. Genet. 34 (2003) 267–273.
[29] M.E. Patti, A.J. Butte, S. Crunkhorn, K. Cusi, R. Berria, S. Kashyap, Y. Miyazaki, I. Kohane, M. Costello, R. Saccone, et al., Coordinated reduction of genes of oxidative metabolism in humans with insulin resistance and diabetes: potential role of PGC1 and NRF1, Proc. Natl. Acad. Sci. U. S. A. 100 (2003) 8466–8471.
[30] R.C. Meex, V.B. Schrauwen-Hinderling, E. Moonen-Kornips, G. Schaart, M. Mensink, E. Phielix, T. van de Weijer, J.P. Sels, P. Schrauwen, M.K. Hesselink, Restoration of muscle mitochondrial function and metabolic flexibility in type 2 diabetes by exercise training is paralleled by increased myocellular fat storage and improved insulin sensitivity, Diabetes 59 (2010) 572–579.
[31] E.V. Menshikova, V.B. Ritov, F.G. Toledo, R.E. Ferrell, B.H. Goodpaster, D.E. Kelley, Effects of weight loss and physical activity on skeletal muscle mitochondrial function in obesity, Am. J. Physiol. Endocrinol. Metab. 288 (2005) E818–E825.
[32] M. Lagouge, C. Argmann, Z. Gerhart-Hines, H. Meziane, C. Lerin, F. Daussin, N. Messadeq, J. Milne, P. Lambert, P. Elliott, et al., Resveratrol improves mitochondrial function and protects against metabolic disease by activating SIRT1 and PGC-1alpha, Cell 127 (2006) 1109–1122.
[33] L.L. Chen, H.H. Zhang, J. Zheng, X. Hu, W. Kong, D. Hu, S.X. Wang, P. Zhang, Resveratrol attenuates high-fat diet-induced insulin resistance by influencing skeletal muscle lipid transport and subsarcolemmal mitochondrial beta-oxidation, Metabolism 60 (2011) 1598–1609.
[34] K.J. Pearson, J.A. Baur, K.N. Lewis, L. Peshkin, N.L. Price, N. Labinskyy, W.R. Swindell, D. Kamara, R.K. Minor, E. Perez, et al., Resveratrol delays age-related deterioration and mimics transcriptional aspects of dietary restriction without extending life span, Cell Metab. 8 (2008) 157–168.
[35] S. Kim, Y. Jin, Y. Choi, T. Park, Resveratrol exerts anti-obesity effects via mechanisms involving down-regulation of adipogenic and inflammatory processes in mice, Biochem. Pharmacol. 81 (2011) 1343–1351.
[36] A. Dal-Pan, S. Blanc, F. Aujard, Resveratrol suppresses body mass gain in a seasonal non-human primate model of obesity, BMC Physiol. 10 (2010) 11.
[37] A. Dal-Pan, J. Terrien, F. Pifferi, R. Botalla, I. Hardy, J. Marchal, A. Zahariev, I. Chery, P. Zizzari, M. Perret, et al., Caloric restriction or resveratrol supplementation and ageing in a non-human primate: first-year outcome of the RESTRIKAL study in *Microcebus murinus*, Age (Dordr) 33 (2011) 15–31.
[38] B. Cannon, J. Nedergaard, Brown adipose tissue: function and physiological significance, Physiol. Rev. 84 (2004) 277–359.
[39] J.M. Andrade, A.C. Frade, J.B. Guimaraes, K.M. Freitas, M.T. Lopes, A.L. Guimaraes, A.M. de Paula, C.C. Coimbra, S.H. Santos, Resveratrol increases brown adipose tissue thermogenesis markers by increasing SIRT1 and energy expenditure and decreasing fat accumulation in adipose tissue of mice fed a standard diet, Eur. J. Nutr. 53 (2014) 1503–1510.
[40] G. Alberdi, V.M. Rodriguez, J. Miranda, M.T. Macarulla, I. Churruca, M.P. Portillo, Thermogenesis is involved in the body-fat lowering effects of resveratrol in rats, Food Chem. 141 (2013) 1530–1535.
[41] Y.H. Tseng, E. Kokkotou, T.J. Schulz, T.L. Huang, J.N. Winnay, C.M. Taniguchi, T.T. Tran, R. Suzuki, D.O. Espinoza, Y. Yamamoto, et al., New role of bone morphogenetic protein 7 in brown adipogenesis and energy expenditure, Nature 454 (2008) 1000–1004.
[42] K.A. Virtanen, M.E. Lidell, J. Orava, M. Heglind, R. Westergren, T. Niemi, M. Taittonen, J. Laine, N.J. Savisto, S. Enerback, et al., Functional brown adipose tissue in healthy adults, N. Engl. J. Med. 360 (2009) 1518–1525.
[43] J.K. Bhatt, S. Thomas, M.J. Nanjan, Resveratrol supplementation improves glycemic control in type 2 diabetes mellitus, Nutr. Res. 32 (2012) 537–541.

[44] J.P. Crandall, V. Oram, G. Trandafirescu, M. Reid, P. Kishore, M. Hawkins, H.W. Cohen, N. Barzilai, Pilot study of resveratrol in older adults with impaired glucose tolerance, J. Gerontol. A Biol. Sci. Med. Sci. 67 (2012) 1307–1312.
[45] M.M. Poulsen, P.F. Vestergaard, B.F. Clasen, Y. Radko, L.P. Christensen, H. Stodkilde-Jorgensen, N. Moller, N. Jessen, S.B. Pedersen, J.O. Jorgensen, High-dose resveratrol supplementation in obese men: an investigator-initiated, randomized, placebo-controlled clinical trial of substrate metabolism, insulin sensitivity, and body composition, Diabetes 62 (2013) 1186–1195.
[46] A. Movahed, I. Nabipour, X. Lieben Louis, S.J. Thandapilly, L. Yu, M. Kalantarhormozi, S.J. Rekabpour, T. Netticadan, Antihyperglycemic effects of short term resveratrol supplementation in type 2 diabetic patients, Evid. Based Complement. Alternat. Med. 2013 (2013) 851267.
[47] L. Rivera, R. Moron, A. Zarzuelo, M. Galisteo, Long-term resveratrol administration reduces metabolic disturbances and lowers blood pressure in obese Zucker rats, Biochem. Pharmacol. 77 (2009) 1053–1063.
[48] J. Shang, L.L. Chen, F.X. Xiao, H. Sun, H.C. Ding, H. Xiao, Resveratrol improves non-alcoholic fatty liver disease by activating AMP-activated protein kinase, Acta Pharmacol. Sin. 29 (2008) 698–706.
[49] W. Kang, H.J. Hong, J. Guan, D.G. Kim, E.J. Yang, G. Koh, D. Park, C.H. Han, Y.J. Lee, D.H. Lee, Resveratrol improves insulin signaling in a tissue-specific manner under insulin-resistant conditions only: in vitro and in vivo experiments in rodents, Metabolism 61 (2012) 424–433.
[50] B.T. Jeon, E.A. Jeong, H.J. Shin, Y. Lee, D.H. Lee, H.J. Kim, S.S. Kang, G.J. Cho, W.S. Choi, G.S. Roh, Resveratrol attenuates obesity-associated peripheral and central inflammation and improves memory deficit in mice fed a high-fat diet, Diabetes 61 (2012) 1444–1454.
[51] J.L. Barger, T. Kayo, J.M. Vann, E.B. Arias, J. Wang, T.A. Hacker, Y. Wang, D. Raederstorff, J.D. Morrow, C. Leeuwenburgh, et al., A low dose of dietary resveratrol partially mimics caloric restriction and retards aging parameters in mice, PLoS One 3 (2008) e2264.
[52] C. Sun, F. Zhang, X. Ge, T. Yan, X. Chen, X. Shi, Q. Zhai, SIRT1 improves insulin sensitivity under insulin-resistant conditions by repressing PTP1B, Cell Metab. 6 (2007) 307–319.
[53] J. Marchal, S. Blanc, J. Epelbaum, F. Aujard, F. Pifferi, Effects of chronic calorie restriction or dietary resveratrol supplementation on insulin sensitivity markers in a primate, *Microcebus murinus*, PLoS One 7 (2012) e34289.
[54] Y. Jimenez-Gomez, J.A. Mattison, K.J. Pearson, A. Martin-Montalvo, H.H. Palacios, A.M. Sossong, T.M. Ward, C.M. Younts, K. Lewis, J.S. Allard, et al., Resveratrol improves adipose insulin signaling and reduces the inflammatory response in adipose tissue of rhesus monkeys on high-fat, high-sugar diet, Cell Metab. 18 (2013) 533–545.
[55] V.S. Chachay, G.A. Macdonald, J.H. Martin, J.P. Whitehead, T.M. O'Moore-Sulliva, P. Lee, M. Franklin, K. Klein, P.J. Taylor, M. Ferguson, et al., Resveratrol does not benefit patients with nonalcoholic fatty liver disease, Clin. Gastroenterol. Hepatol. 12 (2014) 2092–2103.
[56] S. Dash, C. Xiao, C. Morgantini, L. Szeto, G.F. Lewis, High-dose resveratrol treatment for 2 weeks inhibits intestinal and hepatic lipoprotein production in overweight/obese men, Arterioscler. Thromb. Vasc. Biol. 33 (2013) 2895–2901.
[57] K. Magyar, R. Halmosi, A. Palfi, G. Feher, L. Czopf, A. Fulop, I. Battyany, B. Sumegi, K. Toth, E. Szabados, Cardioprotection by resveratrol: a human clinical trial in patients with stable coronary artery disease, Clin. Hemorheol. Microcirc. 50 (2012) 179–187.
[58] P.J. Elliott, S. Walpole, L. Morelli, P.D. Lambert, W. Lunsmann, C.H. Westphal, S. Lavu, Resveratrol/SRT-501, Drugs Future 34 (2009).
[59] P. Brasnyo, G.A. Molnar, M. Mohas, L. Marko, B. Laczy, J. Cseh, E. Mikolas, I.A. Szijarto, A. Merei, R. Halmai, et al., Resveratrol improves insulin sensitivity, reduces oxidative stress and activates the Akt pathway in type 2 diabetic patients, Br. J. Nutr. 106 (2011) 383–389.
[60] N.A. van Herpen, V.B. Schrauwen-Hinderling, G. Schaart, R.P. Mensink, P. Schrauwen, Three weeks on a high-fat diet increases intrahepatic lipid accumulation and decreases metabolic flexibility in healthy overweight men, J. Clin. Endocrinol. Metab. 96 (2011) E691–E695.
[61] WGO, Nonalcoholic fatty liver disease and nonalcoholic steatohepatitis, global guideline, WGO Global Guidelines, 2012, pp. 1–29.
[62] A. Purushotham, T.T. Schug, Q. Xu, S. Surapureddi, X. Guo, X. Li, Hepatocyte-specific deletion of SIRT1 alters fatty acid metabolism and results in hepatic steatosis and inflammation, Cell Metab. 9 (2009) 327–338.
[63] J.T. Rodgers, P. Puigserver, Fasting-dependent glucose and lipid metabolic response through hepatic sirtuin 1, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 12861–12866.
[64] B. Ponugoti, D.H. Kim, Z. Xiao, Z. Smith, J. Miao, M. Zang, S.Y. Wu, C.M. Chiang, T.D. Veenstra, J.K. Kemper, SIRT1 deacetylates and inhibits SREBP-1C activity in regulation of hepatic lipid metabolism, J. Biol. Chem. 285 (2010) 33959–33970.
[65] A.K. Walker, F. Yang, K. Jiang, J.Y. Ji, J.L. Watts, A. Purushotham, O. Boss, M.L. Hirsch, S. Ribich, J.J. Smith, et al., Conserved role of SIRT1 orthologs in fasting-dependent inhibition of the lipid/cholesterol regulator SREBP, Genes Dev. 24 (2010) 1403–1417.
[66] G. Alberdi, V.M. Rodriguez, M.T. Macarulla, J. Miranda, I. Churruca, M.P. Portillo, Hepatic lipid metabolic pathways modified by resveratrol in rats fed an obesogenic diet, Nutrition 29 (2013) 562–567.
[67] M.M. Poulsen, J.O. Larsen, S. Hamilton-Dutoit, B.F. Clasen, N. Jessen, S.K. Paulsen, T.N. Kjaer, B. Richelsen, S.B. Pedersen, Resveratrol up-regulates hepatic uncoupling protein 2 and prevents development of nonalcoholic fatty liver disease in rats fed a high-fat diet, Nutr. Res. 32 (2012) 701–708.
[68] S. Heeboll, K.L. Thomsen, S.B. Pedersen, H. Vilstrup, J. George, H. Gronbaek, Effects of resveratrol in experimental and clinical non-alcoholic fatty liver disease, World J. Hepatol. 6 (2014) 188–198.
[69] P. Xin, H. Han, D. Gao, W. Cui, X. Yang, C. Ying, X. Sun, L. Hao, Alleviative effects of resveratrol on nonalcoholic fatty liver disease are associated with up regulation of hepatic low density lipoprotein receptor and scavenger receptor class B type I gene expressions in rats, Food Chem. Toxicol. 52 (2013) 12–18.
[70] I.J. Cho, J.Y. Ahn, S. Kim, M.S. Choi, T.Y. Ha, Resveratrol attenuates the expression of HMG-CoA reductase mRNA in hamsters, Biochem. Biophys. Res. Commun. 367 (2008) 190–194.
[71] M. Zhou, S. Wang, A. Zhao, K. Wang, Z. Fan, H. Yang, W. Liao, S. Bao, L. Zhao, Y. Zhang, et al., Transcriptomic and metabonomic profiling reveal synergistic effects of quercetin and resveratrol supplementation in high fat diet fed mice, J. Proteome Res. 11 (2012) 4961–4971.
[72] S.J. Cho, U.J. Jung, M.S. Choi, Differential effects of low-dose resveratrol on adiposity and hepatic steatosis in diet-induced obese mice, Br. J. Nutr. 108 (2012) 2166–2175.
[73] Q. Chen, E. Wang, L. Ma, P. Zhai, Dietary resveratrol increases the expression of hepatic 7alpha-hydroxylase and ameliorates hypercholesterolemia in high-fat fed C57BL/6 J mice, Lipids Health Dis. 11 (2012) 56.
[74] Y. Bai, Q.Q. Mao, J. Qin, X.Y. Zheng, Y.B. Wang, K. Yang, H.F. Shen, L.P. Xie, Resveratrol induces apoptosis and cell cycle arrest of human T24 bladder cancer cells in vitro and inhibits tumor growth in vivo, Cancer Sci. 101 (2010) 488–493.
[75] G.M. Do, U.J. Jung, H.J. Park, E.Y. Kwon, S.M. Jeon, R.A. McGregor, M.S. Choi, Resveratrol ameliorates diabetes-related metabolic changes via activation of AMP-activated protein kinase and its downstream targets in db/db mice, Mol. Nutr. Food Res. 56 (2012) 1282–1291.
[76] J. Ahn, I. Cho, S. Kim, D. Kwon, T. Ha, Dietary resveratrol alters lipid metabolism-related gene expression of mice on an atherogenic diet, J. Hepatol. 49 (2008) 1019–1028.
[77] S. Gomez-Zorita, A. Fernandez-Quintela, M.T. Macarulla, L. Aguirre, E. Hijona, L. Bujanda, F. Milagro, J.A. Martinez, M.P. Portillo, Resveratrol attenuates steatosis in obese Zucker rats by decreasing fatty acid availability and reducing oxidative stress, Br. J. Nutr. 107 (2012) 202–210.
[78] J.G. Franco, P.C. Lisboa, N.S. Lima, T.A. Amaral, N. Peixoto-Silva, A.C. Resende, E. Oliveira, M.C. Passos, E.G. Moura, Resveratrol attenuates oxidative stress and prevents steatosis and hypertension in obese rats programmed by early weaning, J. Nutr. Biochem. 24 (2013) 960–966.
[79] M. Gao, D. Liu, Resveratrol suppresses T0901317-induced hepatic fat accumulation in mice, AAPS J. 15 (2013) 744–752.
[80] L. Bujanda, E. Hijona, M. Larzabal, M. Beraza, P. Aldazabal, N. Garcia-Urkia, C. Sarasqueta, A. Cosme, B. Irastorza, A. Gonzalez, et al., Resveratrol inhibits nonalcoholic fatty liver disease in rats, BMC Gastroenterol. 8 (2008) 40.
[81] L. Li, J. Hai, Z. Li, Y. Zhang, H. Peng, K. Li, X. Weng, Resveratrol modulates autophagy and NF-kappaB activity in a murine model for treating non-alcoholic fatty liver disease, Food Chem. Toxicol. 63 (2014) 166–173.
[82] E. Tauriainen, M. Luostarinen, E. Martonen, P. Finckenberg, M. Kovalainen, A. Huotari, K.H. Herzig, A. Lecklin, E. Mervaala, Distinct effects of calorie restriction and resveratrol on diet-induced obesity and fatty liver formation, J. Nutr. Metab. 2011 (2011) 525094.
[83] S.R. Farmer, Obesity: be cool, lose weight, Nature 458 (2009) 839–840.